4-2016

Investigating Dispersal of Red Oak (Quercus rubra) Acorns by the Eastern Gray Squirrel (Sciurus carolinensis)

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Investigating Dispersal of Red Oak (*Quercus rubra*) Acorns by the Eastern Gray Squirrel (*Sciurus carolinensis*)

A thesis submitted in partial fulfillment of the requirement
for the degree of Bachelor of Science in Biology from
The College of William and Mary

by

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Accepted for Honors
(Honors, High Honors, Highest Honors)

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April 19, 2016

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Introduction

One important relationship in deciduous forest ecology is between granivorous animals and plants (Steele et al. 2011). Granivores act as a predatory pressure on seed plants by consuming seeds, but some granivores also contribute to seed germination by dispersing seeds. Depending on the seed predator and plants involved, their specific relationship would be determined to be mutualistic if the plant experiences more recruitment in the presence of seed predators than in their absence (Zwolak and Crone 2012, Lichti et al. 2015). For instance, in the absence of yellow pine chipmunks, Jeffrey pine seeds are left on the soil surface where the seeds cannot germinate (Vander Wall 1992). On the other hand, the tropical tree *Gustavia superba* can successfully germinate under a wide variety of conditions (Sork 1985), so it is unlikely that seed-dispersers can facilitate its recruitment (Zwolak and Crone 2012).

Directed dispersal can strengthen the granivore-seed mutualism. Under directed dispersal, a dispersing agent non-randomly transports seeds to microsites well suited for plant establishment and growth (Howe and Smallwood 1982, Wenny 2001). Although initially posited to be rare (Howe 1986), reports of directed dispersal increased as researchers further investigated the behaviors of animal dispersers in various systems (Wenny and Levey 1998, Pearson and Theimer 2004, Briggs et al. 2009, Yi et al. 2013, Gallegos et al. 2014, Haurez et al. 2016). Wenny (2001) suggests that a poor understanding of seed and seedling shadows generated by dispersing animals contributes to an underappreciation for the prevalence of directed dispersal. Additionally, Wenny (2001) argues that researchers may conflate the benefits of dispersal attributable to directed dispersal with other benefits of dispersal, namely colonization (where seeds are dispersed towards previously uninhabited sites) and escape from density dependent mortality (including low germination rates and high predation and infection rates associated with sites beneath the parent tree). Therefore, ecologists are encouraged to scrutinize the dispersal preferences of animal dispersers and evaluate possible ramifications on dispersed plants.

Several recent studies investigated directed dispersal by rodents. For example, Briggs et al. (2009) found that Jeffrey pine (*Pinus jeffreyi*) seeds produced more seedlings when buried at depths typical of rodent caches. Pearson and Theimer (2004) found that pinyon mice (*Peromyscus truei*) and brush mice (*P. boylii*) cached more seeds in fine particle soil near rocks. These sites were associated with greater pinyon seed germination and higher water content. On the other hand, Iida (2006) found that seeds of the deciduous oak *Quercus serrata* that were dispersed by wood mice had lower survival rates. These studies demonstrate the need for detailed behavioral studies that further understanding on how disperser preferences coincide, or fail to coincide, with the needs of plants.

While it is reasonable to suspect that closely related species can behave similarly, differing climates can produce significantly divergent ecologies (Vander Wall and Jenkins 2011).

For instance, Vander Wall and Jenkins (2011) note that the yellow pine chipmunk (*Tamias amoneus*) attempts to avoid seed pilferage by scatter hoarding seeds, defined by storing one or a few seeds in caches spread over a wide area (Vander Wall 1990). On the other hand, the closely related eastern chipmunk (*Tamias striatus*) larder hoards seeds by storing seeds in a central location where it can defend them against pilferers (Vander Wall 1990). It is suggested that scatter hoarding is favored in dry western environments because olfactory detection of buried seeds is moisture dependent, therefore, seeds can be safely scattered with a low incidence of pilferage (Vander Wall 1998). On the other hand, in the mesic deciduous forests of eastern North America, where buried seeds may be more easily detected and pilfered, larder hoarding is more effective at preventing pilferage (Vander Wall and Jenkins 2011). Ecologists are therefore posed with a question: Do conditions in a particular region prompt animal dispersers to behave in a particular manner? For instance, Yi et al. (2013) observed that Siberian chipmunks (*Tamias sibiricus*) dispersed the acorns of a white oak, *Quercus mongolica*, to sites with high soil water content. While this may be true of rodents in northeastern China, with an average annual precipitation of 650 mm, can the same be said of rodents in the eastern deciduous United States, which can have upwards of 1000 mm of annual precipitation? There is merit, therefore, in conducting disperser-behavior studies on seed-dispersing animals in novel environments, while considering the local and regional effects that may be influencing disperser activity.

We tracked cache sites made by the eastern gray squirrel, *Sciurus carolinensis*, when caching northern red oak (*Quercus rubra*) acorns in an eastern deciduous forest. The seed dispersal behavior of the eastern gray squirrel has been studied extensively (Steele et al. 2001, Steele et al. 2011, Steele et al. 2014, Steele et al. 2015); however, whether the eastern gray squirrel transports seeds to sites favorable for germination has not been specifically examined. We were then able to compare a suite of microsite characteristics at cache sites with characteristics at randomly-chosen sites. This allowed us to predict whether any of the measured characteristics would dictate cache site selection by squirrels. We then planted red oak, white oak (*Quercus alba*), and chestnut (*Castanea dentata*) seeds at cache and random sites. By evaluating rates of seedling emergence in the spring, we will be able to evaluate whether squirrels were directly dispersing seeds to favorable microsite conditions.

Methods

Site Description

Seed stations were established at four sites in the College Woods in Williamsburg, Virginia. The College Woods, also known as Matoaka Woods, is an oak-dominated hardwood forest located on the campus of the College of William and Mary (Kribel et al. 2011). Of the oaks (*Quercus* spp.), white oak (*Q. alba*) is the most common. Other canopy trees include beech (*Fagus grandifolia*), tuliptree (*Liriodendron tulipifera*), and hickories (*Carya* spp.). Species beneath the canopy include holly (*Ilex opaca*), red maple (*Acer rubrum*), dogwood (*Cornus florida*), sourwood (*Oxydendron arboreum*), and blackgum (*Nyssa sylvatica*).

Stations 1 and 2 were located behind the William and Mary Population and Community Ecology Lab. This area was cleared in 1950, and thus, is populated by young trees that grew in the absence of an oak canopy. Longleaf pine (*Pinus palustris*) dominates the canopy.

Seeds in the College Woods experience dispersal and consumption at the hands of two scatterhoarding rodents: the eastern gray squirrel (*Sciurus carolinensis*), and the white footed mouse (*Peromyscus leucopus*).

Plant Species Descriptions

Northern red oak (*Quercus rubra*) is a deciduous canopy tree that grows up to 30 m. Of the three study species, red oak acorns contain the highest lipid and energy content, and have the highest tannin content (Lichti et al. 2014, Steele et al. 2001). Additionally, red oak acorns undergo the longest period of dormancy before germinating. Lichti et al. (2014) found that rodents tend to cache imperishable, unpalatable seeds such as red oak, and consume perishable, palatable seeds such as white oak.

White oak (*Quercus alba*) is a deciduous canopy tree that grows up to 25 m. White oak acorns exhibit relatively low lipid, energy, and tannin levels and germinate immediately upon dropping from the tree (Lichti et al. 2014, Steele et al. 2001). Therefore, when presented white oak and red oak acorns, scatterhoarding rodents are generally expected to consume white oak and cache red oak.

American chestnut (*Castanea dentata*) is a large tree that has previously been known to grow to 30 m. Chestnut seeds do not express tannin activity, and have an intermediate energy yield and dormancy period (Lichti et al. 2014). Once a dominant tree of the deciduous forests of the United States, American chestnut has been largely replaced by oaks as a result of the introduction of the fungal pathogen chestnut blight (*Cryphonectria parasitica*) (Blythe et al. 2015).

Seed Collection and Processing

Seed collection spanned October 10th to October 24th 2015. Seeds were sourced from under the canopies of 4 northern red oak and 7 white oak trees located in Colonial Williamsburg. B3F2 chestnuts were provided by the breeding program of the Maryland chapter of the American Chestnut Foundation. Seeds were visually inspected for weevil infestation and other signs of unviability before being float tested. Seeds that passed screening were preserved in Ziploc bags surrounded by moist soil. Until they were further processed or used in the field, seeds were kept refrigerated.

In order to track squirrel cache locations, 1000 northern red oak acorns were tagged with pink tagging. Acorns were drilled with a 1.59 mm (1/16 in.) drill bit near the base of the seed so as not to damage the embryo. Acorns were then visually screened for cracks and other artifacts of the drilling process. Acorns were threaded with 0.95 mm (0.0375 in.) wire that was attached to pink tags. Acorns were then returned to moist soil and refrigerated until needed.

Squirrel Preference Procedure

At four stations, special exclusion cages were erected in order to prevent white tailed deer, raccoons, and other large granivorous animals from exploiting seed stations, while allowing grey squirrels access. The tops of the cages were constructed from a 1x1 m^2 section of chicken wire. The cage tops were propped off the ground by four PVC pipes, each 0.5 m in length with approximately 0.2 meters of the length of the pipe pounded into the soil. At the top of each pipe was a small hole through which a zip tie was threaded. These zip ties were used to secure the PVC pipes to the top of the cages. Additional chicken wire was added to form the sides of the cages. The cage sides were secured to the top of the cage with zip ties, and the bottom of the cages were fastened to the ground using field staples. Rectangular holes were cut out of the center-bottom of each of the four sides of the cage to form holes approximately 15 x 20 cm for the squirrels to pass through. Camera footage from preliminary research demonstrated that squirrels were able to enter and exit the cage through these holes. There was no evidence that the cages deterred the squirrels from exploiting the site.

Mixed squirrel feed was placed at stations 1 and 2 on November 7th. This was done so that squirrels would become acquainted with the sites as sources of food. On November 10th, the squirrel feed was removed and 250 tagged red oak acorns were placed at each of the sites. The sites were then left unattended until November 14th. Preliminary research demonstrated that squirrels tend to fully exploit such sites in three to four days. The same methods were performed at stations 3 and 4, with feed being placed on November 14th and acorns being placed on November 17th, and left unattended until November 21st. From the time of feed being set out to data being collected, sites were monitored with hunting cameras in order to characterize the fauna that exploited the sites.

Cache sites were located four days after tagged seeds were set out. A total of 1.5 person hours was allocated to search a roughly 30-m radius area at each seed station. Footage from the hunting cameras confirmed that only squirrels and raccoons had access to the seeds. Raccoons consume seeds but do not bury them, therefore, any burying of the seeds was attributed to squirrels. A tagged seed was determined to be a squirrel-chosen cache site if the seed was at least partially below the soil surface.

A total of 164 cache sites were found among the 4 sites. At each cache site, burial depth, soil moisture, distance to nearest tree (with diameter at breast height at least 10 cm), understory cover, and overstory cover was determined. Burial depth was taken from the surface of the soil to the bottom of the cache. Soil moisture was taken using a soil moisture meter. Overstory cover was determined with a densiometer. Understory was characterized by placing 1x1 m^2 PVC quadrats at each site, centered on the cache. For each quadrat, percent cover of leaf/needle litter, woody debris, herbaceous plants, and woody plants was estimated.

Each cache site was paired with a random site. Random sites were determined from random distances from the seed station of 0-30 m and random headings of 0-360 degrees, with 0 being north. At each random site, soil moisture, distance to tree, understory characteristics, and overstory cover were recorded, with the same methods as the cache sites.

Directed Dispersal Procedure

One red oak, one white oak, and one chestnut seed was planted at each of the cache sites located in stations 1 and 2, and 40 sites at station 3. These seeds were also planted at the random coordinates located at stations 1 and 2, and 40 random sites at station 3. Each seed cache was buried to a depth of approximately 2.5 cm.

Half of the squirrel-chosen cache sites and half of the random sites were protected with exclusion cylinders. These cylinders were designed to exclude granivorous rodents above and belowground. A 10 cm diameter PVC pipe 15 cm long was pounded into the soil, leaving approximately 1 cm aboveground. Hardware cloth 10 cm in height was wrapped around the aboveground portion of the PVC pipe and attached with zip ties threaded through four holes drilled into the PVC. The top of the hardware cloth was attached to a square section of hardware cloth, in dimensions of 10 x 10 cm, with additional zip ties. However, due to time constraints, seeds to be protected at station 3 were protected with a cylinder of hardware cloth affixed to the soil with landscape staples.

The other half of the cache sites and random sites were left unprotected in order to compare pilferage rates between squirrel chosen sites and random sites.

All artificial caches will be left unattended until late April. When revisited, artificial caches behind the School of Law and in the College Woods will be recovered and their status (emerged, dormant, dead, missing) will be recorded.

Data Analysis

All statistics were performed in R. Differences in soil moisture, distance to nearest tree, and canopy cover at cache sites and random sites were determined by applying a linear mixed effects model to each response variable using the lmer function of the lme4 package. For each linear model, treatment (cache or random) was designated as a fixed effect and station (1, 2, or 3) was designated as a random effect. Differences in understory composition were similarly examined with linear models, however, the distribution of the residuals of these models raised concerns about meeting the assumptions of a linear model. Therefore, understory characteristics at cache sites and random sites at station 3 were compared with the non-parametric Kolmogorov-Smirnov test.

Correlations between microsite characteristics were determined using the corr.test function of the psych package. We also performed a discriminant function analysis (DFA) in an attempt to generate a function that can successfully predict whether a site is a cache or random site based on a combination of its microsite characteristics. If successful, the linear discriminant generated by the DFA would assign each site a value along the discriminant function such that cache sites have similar values and random sites have similar values with little overlap between the cache and random sites. DFAs were performed using the lda function of the MASS package.

Results

A total of 164 squirrel caches were located (Station 1: $n = 30$; Station 2: $n = 8$; Station 3: $n = 126$; Station 4: $n = 0$). At Station 3, a total of 132 tags out of 250 were relocated, for a recovery rate of 53.6%.

A total of 148 random sites were located (Station 1: $n = 29$; Station 2: $n = 15$; Station 3: $n = 104$).

Mean squirrel cache depth was 2.45 cm ($s = 0.901$).

Cache Site Characteristics

Squirrel caches were drier (having lower percent moisture) than random locations ($\bar{x}_C = 14.3,\ \bar{x}_R = 16.42,\ t = 2.87,\ df = 308,\ P = 0.00439$) (see Figures 1 and 2). Additionally, squirrel caches had less percent cover of woody debris around them than in random sites ($\bar{x}_C = 3.546,\ \bar{x}_R = 8.206,\ D = 0.39728,\ P = 4.645 \times 10^{-11}$) (Fig. 3 and 4).

While not significant at $\alpha = 0.05$, squirrel caches may have been located nearer to trees with DBH >= 10 cm ($\bar{x}_C = 1.746\ m,\ \bar{x}_R = 1.986\ m,\ t = 1.878,\ df = 310,\ P = 0.0613$) (Fig. 5 and 6). We observed two positive outlier values for distances to nearest tree at cache sites, defined as values exceeding the third quartile plus 1.5 times the interquartile range. When the same comparison is performed but outlier values are omitted, squirrel cache sites were significantly closer to trees than random sites ($\bar{x}_C = 1.687\ m,\ \bar{x}_R = 1.986\ m,\ t = 2.491,\ df = 308,\ P = 0.0133$).

There was no difference in percent canopy cover between cache and random sites ($\bar{x}_C = 78.12,\ \bar{x}_R = 76.20,\ t = -1.077,\ df = 308,\ P = 0.282179$). There was also no difference in percent cover of leaves and needles around cache and random sites ($\bar{x}_C = 87.39,\ \bar{x}_R = 84.56,\ D = 0.059443,\ P = 0.9469$) There was no difference in percent cover of herbaceous plants around cache sites or random sites. ($\bar{x}_C = 3.227,\ \bar{x}_R = 2.922,\ D = 0.023835,\ P = 1$). Finally, there was no difference in woody vegetation around cache sites compared to random sites ($\bar{x}_C = 3.767,\ \bar{x}_R = 2.270, D = 0.098077,\ P = 0.4446$).

Multivariate Analyses

No significant correlations of large magnitude were found between microsite characteristics (see Table 1). Note that understory characteristics, including percent cover of leaves/needles, wood debris, etc., are not independent from each other and therefore are inherently correlated.

The DFA produced a single linear discriminant to account for the variation in microsite characteristics between cache sites and random sites (Table 2, Figure 7). The DFA appears to indicate that squirrels cache in a subset of random available sites, but choose to not cache in sites with certain characteristics. The linear discriminant is primarily loaded on herbaceous and woody plant cover, despite the fact that neither of these characteristics appear to differ between cache sites and random sites. This may suggest that none of the linear discriminants

on their own have significant predictive power with regards to determining whether a site is suitable for caching.

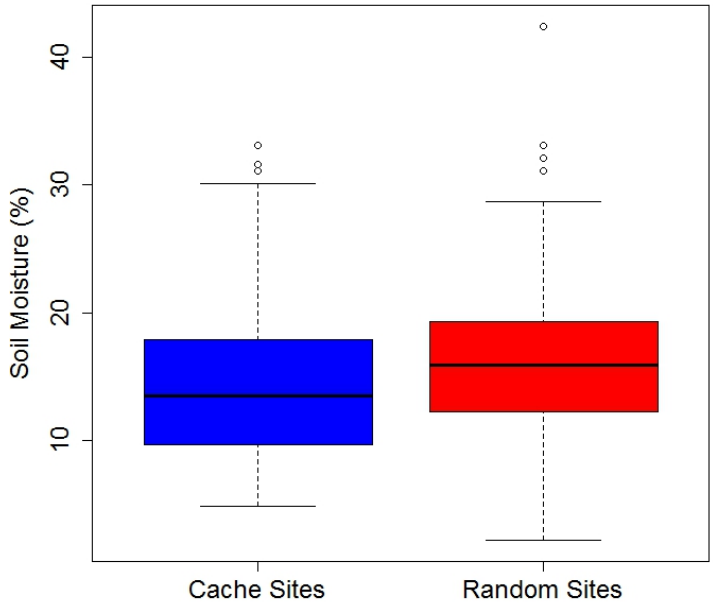

Figure 1. Percent soil moisture at cache sites and random sites. Mean soil moisture at cache sites was lower than mean soil moisture at random sites ($\bar{x}_C = 14.3,\ \bar{x}_R = 16.42,\ t = 2.87,\ df = 308,\ P = 0.00439$).

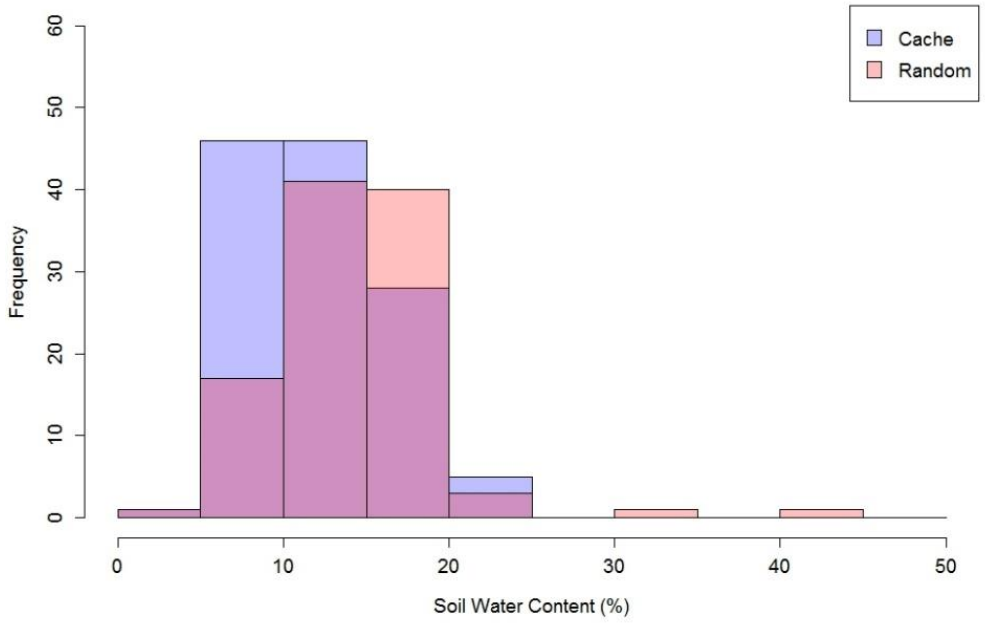

Figure 2. Distributions of percent soil moisture at cache sites (blue) and random sites (red). Purple regions indicate areas of overlap of the two distributions. Mean soil moisture at cache sites was lower than mean soil moisture at random sites ($\bar{x}_C = 14.3,\ \bar{x}_R = 16.42,\ t = 2.87,\ df = 308,\ P = 0.00439$).

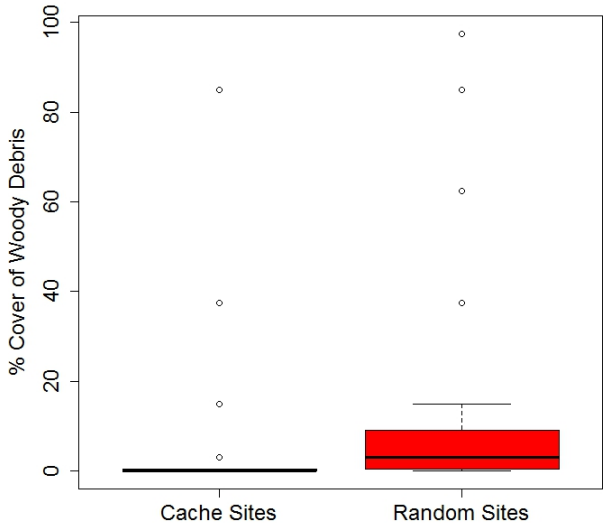

Figure 3. Percent cover of woody debris at cache sites and random sites. Mean wood debris cover at cache sites was lower than mean wood debris cover at random sites ($\bar{x}_C = 3.546,\ \bar{x}_R = 8.206,\ D = 0.39728,\ P = 4.645 \times 10^{-11}$).

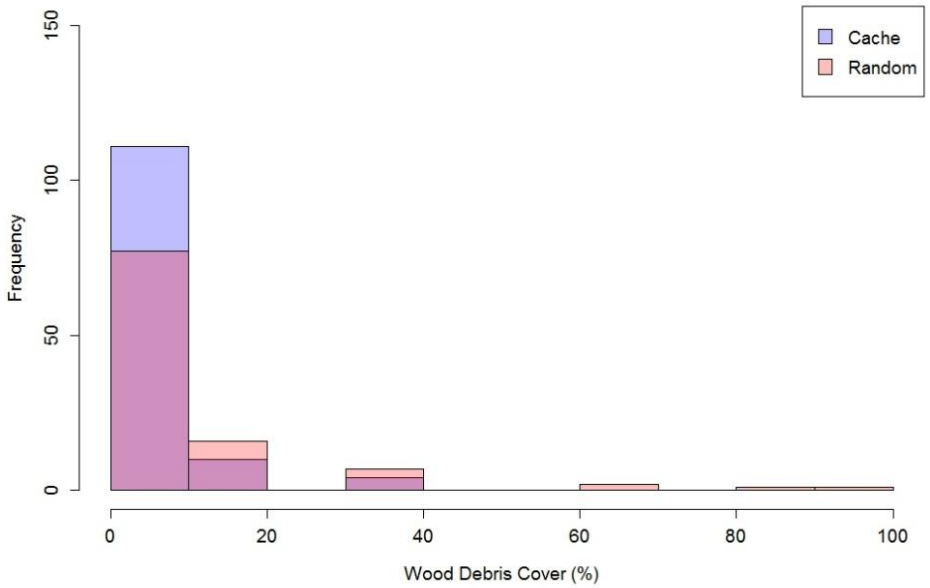

Figure 4. Distributions of percent cover of wood debris around cache sites (blue) and random sites (red). Purple regions indicate areas of overlap of the two distributions. Mean wood debris cover at cache sites was lower than mean wood debris cover at random sites ($\bar{x}_C = 3.546,\ \bar{x}_R = 8.206,\ D = 0.39728,\ P = 4.645 \times 10^{-11}$).

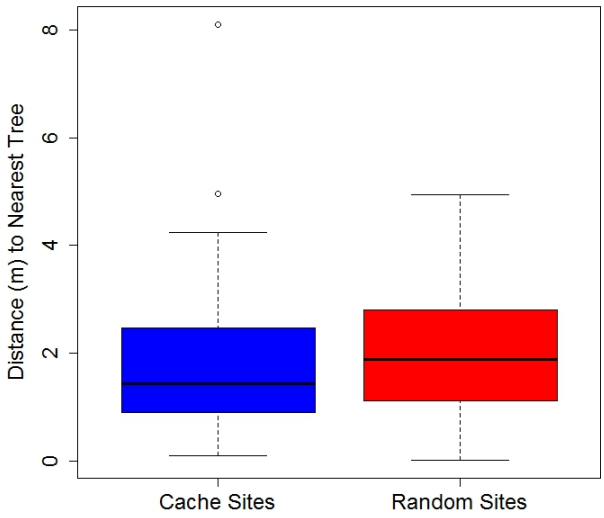

Figure 5. Distance to nearest tree (DBH >= 10 cm) at cache sites and random sites. Mean distance to nearest tree at cache sites was not significantly different than distance to nearest tree at random sites ($\bar{x}_C = 1.746\ m,\ \bar{x}_R = 1.986\ m,\ t = 1.878,\ df = 310,\ P = 0.0613$). However, when two cache site outlier values are excluded from analysis, *mean distance to nearest tree at cache sites was lower than mean distance to nearest tree at random sites* ($\bar{x}_C = 1.687\ m,\ \bar{x}_R = 1.986\ m,\ t = 2.491,\ df = 308,\ P = 0.0133$).

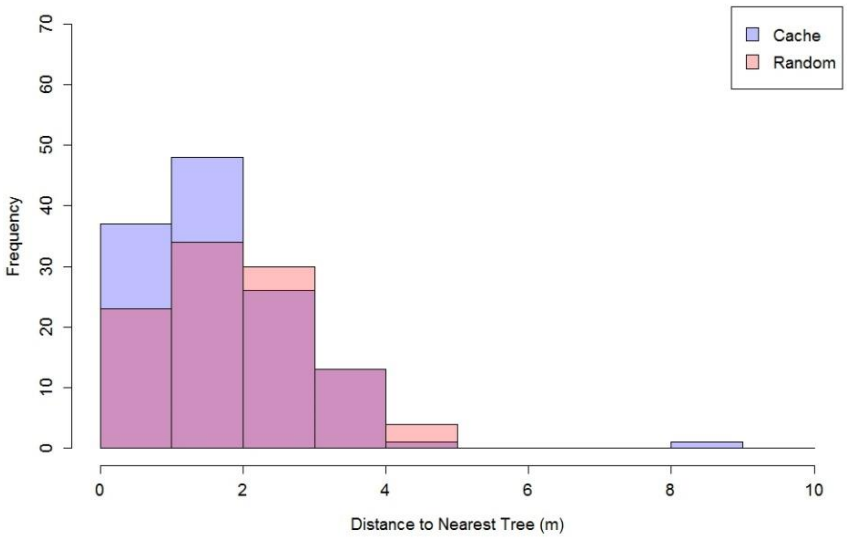

Figure 6. Distributions of distance to nearest tree (DBH >= 10 cm) at cache sites (blue) and random sites (red). Purple regions indicate areas of overlap of the two distributions. Mean distance to nearest tree at cache sites was not significantly different than distance to nearest tree at random sites ($\bar{x}_C = 1.746\,m,\ \bar{x}_R = 1.986\,m,\ t = 1.878,\ df = 310,\ P = 0.0613$). However, when two cache site outlier values are excluded from analysis, *mean distance to nearest tree at cache sites was lower than mean distance to nearest tree at random sites* ($\bar{x}_C = 1.687\,m,\ \bar{x}_R = 1.986\,m,\ t = 2.491,\ df = 308,\ P = 0.0133$).

Corr. Coef.	moisture	treeDistance	densiometer	leaves	wood	herbPlant	woodPlant
moisture	1	0.0893379	**-0.2647573**	**-0.227367**	-0.01752	**0.2959417**	0.0699571
treeDistance	0.089338	1	-0.1167093	-0.063465	**0.171986**	-0.036598	-0.123354
densiometer	-0.26476	-0.1167093	1	**0.245538**	-0.012271	**-0.174519**	0.0205493
leaves	-0.22737	-0.0634652	0.24553776	1	**-0.472511**	**-0.524887**	**-0.358971**
wood	-0.01752	0.17198575	-0.012271	-0.472511	1	-0.016269	-0.105674
herbPlant	0.295942	-0.036598	-0.174519	-0.524887	-0.016269	1	0.023334
woodPlant	0.069957	-0.1233545	0.02054931	-0.358971	-0.105674	0.023334	1

P-Values	moisture	treeDistance	densiometer	leaves	wood	herbPlant	woodPlant
moisture	0	1	**3.72E-05**	**0.000779**	1	**1.89E-06**	1
treeDistance	0.115294	0	0.43661392	1	**0.0304**	1	0.3556212
densiometer	2.19E-06	0.03969217	0	**0.000196**	1	**0.0285921**	1
leaves	5.20E-05	0.26449078	1.23E-05	0	**0**	**0**	**1.30E-09**
wood	0.758268	0.00233845	0.82962076	0	0	1	0.6270396
herbPlant	1.05E-07	0.52020557	0.00204229	0	0.775047	0	1
woodPlant	0.218605	0.0296351	0.71855967	6.86E-11	0.062704	0.68188	0

Table 1. Summary of correlation coefficients between microsite characteristics. P-values above the diagonal are corrected for multiple comparisons. Significant values are in bold. Note that understory values (leaves, wood debris, etc.) are not independent from each other, and thus, comparisons between them are inappropriate.

Coefficients of linear discriminant	
	LD1
Herbaceous Plants	-1.12834276
Woody Plants	0.72974115
Wood Debris	0.23588158
Distance to Nearest Tree	0.20939296
Moisture	0.11398734
Leaf/Needle Cover	0.02411819
Canopy Cover	0.01605908

Table 2. Coefficients of the linear discriminant produced by the Discriminant Function Analysis. Larger magnitude values indicate a greater contribution of each predictor variable in assigning a site (cache or random) a value on the Linear Discriminant. Positive values indicate sites being placed further to the right with increasing value of the predictor, while negative values indicate sites being placed further to the left with increasing value of the predictor (see Figure 7).

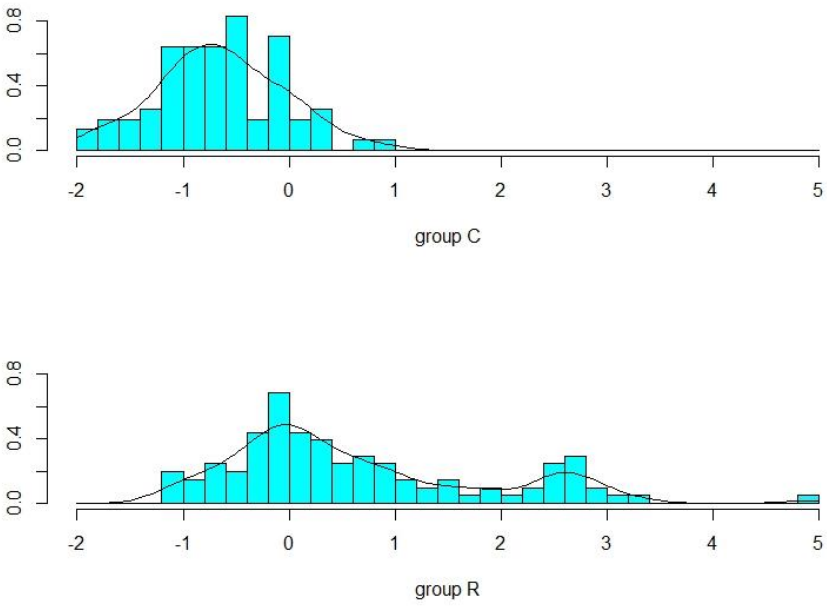

Figure 7. Histograms of each observation along the single discriminant function, grouped by type (C : cache; R : random). There is significant overlap between the values of cache and

random sites, raising doubts as to the ability of the discriminant function to discriminate between cache and random sites with the provided microsite characteristics. However, cache sites appear constrained to linear discriminant values less than 1, with random sites occasionally exceeding 1.

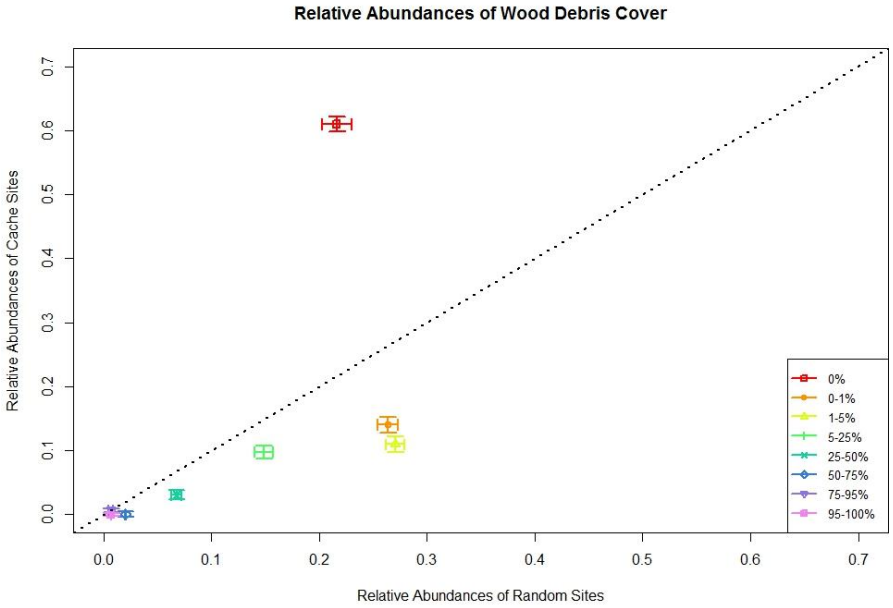

Figure 8. Relative abundancies of wood debris at random sites vs. cache sites. Each point is defined along the x-axis as the proportion of random sites within a cover class out of all random sites measured, and the y-axis as the proportion of cache sites within a cover class out of all cache sites measured. A 1:1 line is plotted to illustrate the null expectation that squirrels exploit cache sites with particular abundances of wood debris in proportion to the available sites that have those abundances of wood debris. Each cover class is represented by a unique symbol and color. Squirrels under-utilize sites with 0-1 and 1-5% wood cover, and greatly over-utilize sites with 0% wood debris.

Discussion

By tracking squirrel cache sites and comparing microsite characteristics from these sites and random sites, we found that eastern gray squirrels prefer to cache red oak acorns in sites with relatively low water content and with low wood debris cover. Additionally, there is some evidence that squirrels may cache red oak acorns at sites closer to trees than random sites. There was no evidence that squirrel cache sites differ from random locations with respect to canopy cover, leaf and needle cover, green plant cover, or woody plant cover.

It is well understood that rodents can more easily detect seeds buried in moist soil via olfactory cues (Vander Wall 1993, Geluso 2005, Downs and Vander Wall 2009). Given that

rodents have a retriever's advantage over conspecific pilferers (see Steele et al. 2011), one might reasonably suspect that rodents would prefer to disperse seeds to wet soils to facilitate recovery. However, we found that eastern gray squirrels selectively scatter-hoard seeds in soils of low water content. This result contrasts with findings by Yi et al. (2013), where Siberian chipmunks directly disperse seeds of a white oak to sites of higher water content. There are a number of reasons why drier cache sites would be preferred. First, as suggested by Yi et al. (2013), the discrepancy in rodent behavior may be explained by differing climates: Annual precipitation in northeastern China averages 650 mm, while annual precipitation in Virginia exceeds 1200 mm. In the relatively drier China climate, small increases in soil moisture may significantly facilitate cache recovery, whereas in the relatively wetter Virginia clime, small changes in soil moisture may not significantly affect cache recovery rates. Second, scatterhoarders may rely on spatial memory, rather than olfaction, to recover their own cached seeds, but use olfactory cues in order to pilfer caches made by other rodents (Vander Wall 2000). Therefore, caching in drier soils may reduce the incidence of cache pilferage. Finally, scatter hoarders may prefer to cache seeds in locations that reduce seed perishability. For instance, Neuschulz et al. (2015) found that nutcrackers preferred to cache seeds in locations with closed canopies and low soil moisture. Seeds cached at these sites had low rates of emergence and therefore perishability.

We have found that squirrel cache sites had less percent cover of woody debris than random sites in the College Woods. In contrast, wood mice have been shown to disperse seeds towards woody debris (Iida 2006, Takahasi et al. 2006). Wood mice utilize the bases of standing trees and fallen branches as habitat and refuge from predators and thus disproportionately disperse sites to these locations. We propose that squirrels are large enough that fallen twigs branches do not offer the substantial protection from predation that twigs and branches may afford mice. Furthermore, high densities of woody debris may interfere with the ability of squirrels to physically access potential cache sites. Therefore, squirrel cache sites would be located in sites on the forest floor with sparse wood debris cover.

We found some evidence that squirrels may disperse seeds towards trees. Dispersing seeds toward trees may be related to managing predation risks. For instance, Steele et al. (2015) found that squirrels perceive sites near the bases of trees as being safer than sites further from trees. However, this was only the case in an open stand setting, and not the case in a natural forest. Furthermore, rather than avoid risky sites, squirrels may seek risky sites as ideal places to cache in order to reduce the incidence of cache pilferage (Steele et al. 2014, Steele et al. 2015). We suggest that the different findings of previous research and our own may be explained by differing predation and pilfering landscapes. If the risk of predation is high and the risk of pilferage is low, squirrels would be incentivized to cache seeds in safe sites without regard for the risk of pilferage. Future studies that characterize the pilferage and predation risk landscapes of the College Woods could provide insight into which pressures drive squirrel behavior.

Discriminant function analysis distinguishing cache and random sites produced a linear discriminant that loaded primarily on herbaceous and woody plant cover followed by wood debris and distance to nearest tree. This is surprising considering that we observed no

difference in plant cover between cache and random sites. A visual representation of the DFA indicates that squirrel caches tend to be located in a subset of random sites with certain characteristics. It is important to note that squirrel cache sites and random sites are not mutually exclusive and that we cannot determine whether a random site may be a suitable cache site for a squirrel in the future. To more specifically test whether squirrels prefer certain site characteristics for cache site selection, one can compare the relative abundances of random sites with various ranges of values for a given microsite characteristic against the relative abundancies of cache sites with those same microsite values (for an example, see Figure 8). For instance, squirrels appear to greatly underutilize sites with no wood debris cover, but overexploit sites with 0-1 and 1-5% cover of wood debris. Behavioral experiments in which squirrels are presented with the option of caching in, for instance, either dry or wet soil, would provide further evidence as to whether squirrels actively seek sites with certain characteristics and whether they avoid others.

After comparing rates of emergence at cache sites and random sites, we will be able to assess whether squirrels are dispersing red oak acorns to sites favorable for germination. In the event that seeds have greater emergence rates at cache sites, we would then be interested in the microsite characteristics at cache sites that are responsible for favorable establishment. We have identified soil moisture, wood debris cover, and distance to nearest tree as characteristics that differ between cache and random sites, but seeds might respond to a characteristic we have not quantified. Linking squirrel cache-site decision-making to the net benefits of seeds transported to these conditions will further illustrate the nature of the interaction between squirrels and the seeds that they disperse.

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